

## WHAT IS CLAIMED IS:

Sub A<sup>1</sup> > 5 1. A peptide nucleic acid conjugate comprising:  
a peptide nucleic acid;  
said peptide nucleic acid having a backbone;

said backbone having an amino end, a carboxyl end, and  
a plurality of amino groups;  
said amino groups each having a tethered nucleobase;  
and a conjugate bound to said peptide nucleic acid either  
directly or through a linking moiety.

10 2. A peptide nucleic acid conjugate of claim 1  
wherein said conjugate is bound through said linking moiety  
to at least one of said backbone, said tether, or said  
nucleobase.

15 3. A peptide nucleic acid conjugate of claim 1  
wherein said conjugate is bound to said backbone.

4. A peptide nucleic acid conjugate of claim 3  
wherein said conjugate is bound to at least one of said  
amino end or said carboxyl end of said backbone.

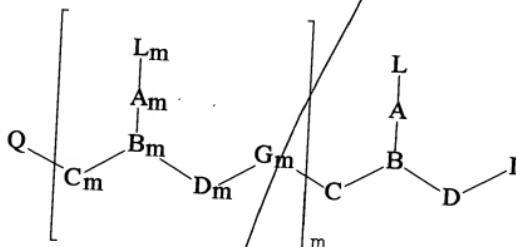
Sub A<sup>2</sup> > 20 5. A peptide nucleic acid conjugate of claim 1  
wherein said conjugate is bound to said nucleobase or said  
tether.

Nielsen et al.  
08/319,411  
f 1 10-6-94

530/350  
536/22.1

6. A peptide nucleic acid conjugate of claim 1 wherein said conjugate is a reporter enzyme, a reporter molecule, a steroid, a carbohydrate, a terpene, a peptide, a protein, an aromatic lipophilic molecule, a non aromatic lipophilic molecule, a phospholipid, an intercalator, a cell receptor binding molecule, a crosslinking agent, a water soluble vitamin, a lipid soluble vitamin, an RNA cleaving complex, a metal chelator, a porphyrin, an alkylator, or a polymeric compound selected from polymeric amines, polymeric glycols and polyethers.

7. A peptide nucleic acid conjugate of the formula:



Sub A<sup>3</sup>

wherein:

m is an integer from 1 to about 50;

L and L<sub>m</sub> independently are R<sup>12</sup>(R<sup>13</sup>)<sub>n</sub>; wherein:

15 R<sup>12</sup> is hydrogen, hydroxy, (C<sub>1</sub>-C<sub>4</sub>) alkanoyl, a

E

5

naturally occurring nucleobase, a non-naturally occurring nucleobase, an aromatic moiety, a DNA intercalator, a nucleobase-binding group, a heterocyclic moiety, a reporter ligand, or a conjugate;

10

provided that at least one of R<sup>12</sup> is a naturally occurring nucleobase, a non-naturally occurring nucleobase, a DNA intercalator, or a nucleobase-binding group;

R<sup>13</sup> is a conjugate; and  
a is 0 or 1;

15

C and C<sub>m</sub> independently are (CR<sup>6</sup>R<sup>7</sup>)<sub>y</sub>; wherein:

R<sup>6</sup> and R<sup>7</sup> independently are hydrogen, a side chain of a naturally occurring alpha amino acid, (C<sub>2</sub>-C<sub>6</sub>) alkyl, aryl, aralkyl, heteroaryl, hydroxy, (C<sub>1</sub>-C<sub>6</sub>) alkoxy, (C<sub>1</sub>-C<sub>6</sub>) alkylthio, a conjugate, NR<sup>3</sup>R<sup>4</sup>, SR<sup>5</sup> or R<sup>6</sup> and R<sup>7</sup> taken together complete an alicyclic or heterocyclic system;

20

wherein R<sup>5</sup> is hydrogen, a conjugate, (C<sub>1</sub>-C<sub>6</sub>) alkyl, hydroxy-, alkoxy-, or alkylthio-substituted (C<sub>1</sub>-C<sub>6</sub>) alkyl; and

25

R<sup>3</sup> and R<sup>4</sup> independently are hydrogen, a conjugate, (C<sub>1</sub>-C<sub>4</sub>) alkyl, hydroxy- or alkoxy- or alkylthio-substituted (C<sub>1</sub>-C<sub>4</sub>) alkyl, hydroxy, alkoxy, alkylthio or amino;

D and D<sub>m</sub> independently are (CR<sup>6</sup>R<sup>7</sup>)<sub>z</sub>;

*Sub A  
cont*

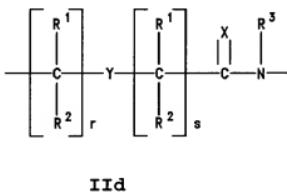
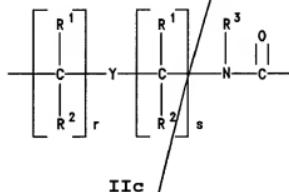
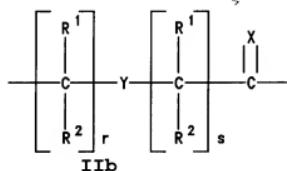
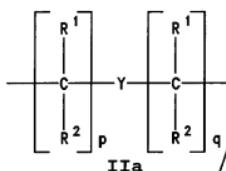
each of  $y$  and  $z$  is zero or an integer from 1 to 10,  
 wherein the sum  $y + z$  is greater than 2 but not more than  
 10;

$G_m$  is independently  $-NR^3CO-$ ,  $-NR^3CS-$ ,  $-NR^3SO-$ , or  
 5  $-NR^3SO_2-$  in either orientation;

each pair of  $A-A_m$  and  $B-B_m$  are selected such that:

- (a)  $A$  or  $A_m$  is a group of formula (IIa), (IIb) or  
 (IIc) and  $B$  or  $B_m$  is N or  $R^3N^+$ ; or  
 (b)  $A$  or  $A_m$  is a group of formula (IID) and  $B$  or  $B_m$  is

10 CH;



E  
wherein:

X is O, S, Se, NR<sup>3</sup>, CH<sub>2</sub> or C(CH<sub>3</sub>)<sub>2</sub>;

Y is a single bond, O, S or NR<sup>4</sup>;

each of p and q is zero or an integer from 1 to 5, the  
5 sum p+q being not more than 10;

each of r and s is zero or an integer from 1 to 5, the  
sum r+s being not more than 10;

R<sup>1</sup> and R<sup>2</sup> independently are hydrogen, (C<sub>1</sub>-C<sub>4</sub>)alkyl,  
hydroxy-substituted (C<sub>1</sub>-C<sub>4</sub>)alkyl, alkoxy-substituted (C<sub>1</sub>-  
10 C<sub>4</sub>)alkyl, alkylthio-substituted (C<sub>1</sub>-C<sub>4</sub>)alkyl, hydroxy,  
alkoxy, alkylthio, amino, halogen or a conjugate;

I is -NR<sup>8</sup>R<sup>9</sup> or -NR<sup>10</sup>C(O)R<sup>11</sup>; wherein:

R<sup>8</sup>, R<sup>9</sup>, R<sup>10</sup> and R<sup>11</sup> independently are hydrogen,  
alkyl, an amino protecting group, a reporter  
15 ligand, an intercalator, a chelator, a peptide, a  
protein, a carbohydrate, a lipid, a steroid, a  
nucleoside, a nucleotide, a nucleotide  
diphosphate, a nucleotide triphosphate, an  
oligonucleotide, an oligonucleoside, a soluble  
20 polymer, a non-soluble polymer or a conjugate;

Q is -CO<sub>2</sub>H, -CO<sub>2</sub>R<sup>8</sup>, -CO<sub>2</sub>R<sup>9</sup>, -CONR<sup>8</sup>R<sup>9</sup>, -SO<sub>3</sub>H, -SO<sub>2</sub>NR<sup>10</sup>R<sup>11</sup> or  
an activated derivative of -CO<sub>2</sub>H or -SO<sub>3</sub>H; and

wherein at least one of R<sup>1</sup>, R<sup>2</sup>, R<sup>3</sup>, R<sup>4</sup>, R<sup>5</sup>, R<sup>6</sup>, R<sup>7</sup>, R<sup>8</sup>,  
25 R<sup>9</sup>, R<sup>10</sup>, R<sup>11</sup>, R<sup>12</sup> and R<sup>13</sup> is a conjugate wherein said conjugate  
is a reporter enzyme, a reporter molecule, a steroid, a

carbohydrate, a terpene, a peptide, a protein, an aromatic lipophilic molecule, a non aromatic lipophilic molecule, a phospholipid, an intercalator, a cell receptor binding molecule, a crosslinking agent, a water soluble vitamin, a lipid soluble vitamin, an RNA cleaving complex, a metal chelator, a porphyrin an alkylator, or a polymeric compound selected from polymeric amines, a polymeric glycols and polyethers; and

wherein said conjugate optionally includes a linking moiety.

8. A peptide nucleic acid conjugate of claim 7  
wherein said conjugate includes a linking moiety.

9. A peptide nucleic acid conjugate of claim 7  
wherein at least one group R<sup>12</sup> is a conjugate.

10. A peptide nucleic acid conjugate of claim 7  
wherein at least one group R<sup>13</sup> is a conjugate.

11. A peptide nucleic acid conjugate of claim 7  
wherein at least one of R<sup>1</sup>, R<sup>2</sup> or R<sup>3</sup> is a conjugate.

12. A peptide nucleic acid conjugate of claim 7  
wherein at least one of said A-A<sub>m</sub> groups include at least one of R<sup>1</sup>, R<sup>2</sup>, and R<sup>3</sup>.

F 13. A peptide nucleic acid conjugate of claim 11,  
wherein at least one of said B-B<sub>m</sub> groups or said G-G<sub>m</sub> groups  
include at least one group R<sup>3</sup>.

B 14. A peptide nucleic acid conjugate of claim 37  
5 wherein at least one of R<sup>8</sup>, R<sup>9</sup>, R<sup>10</sup> and R<sup>11</sup> is a conjugate.

J Sub 15. A peptide nucleic acid conjugate of claim 37  
Sub 36 wherein at least one of said groups Q or I include at least  
one of groups R<sup>8</sup>, R<sup>9</sup>, R<sup>10</sup> and R<sup>11</sup>.

B 16. A peptide nucleic acid conjugate of claim 37  
10 wherein at least one of R<sup>3</sup>, R<sup>4</sup>, R<sup>5</sup>, R<sup>6</sup> and R<sup>7</sup> is a conjugate.

17. A peptide nucleic acid conjugate of claim 16  
wherein at least one of said groups D-D<sub>m</sub>, or C-C<sub>m</sub> include at  
least one of R<sup>3</sup>, R<sup>4</sup>, R<sup>5</sup>, R<sup>6</sup> and R<sup>7</sup>.

B 18. A peptide nucleic acid conjugate of claim 37  
15 wherein m is from 1 to about 200.

B 19. A peptide nucleic acid conjugate of claim 37  
wherein m is from 1 to about 50.

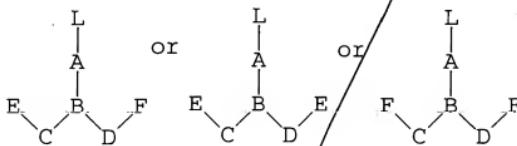
B

*E  
S16  
57*

20. A peptide nucleic acid conjugate of claim 7

wherein m is from 1 to about 20.

21. A compound having one of the following formulas:

*Sub A*  
*57*  
wherein:5       L is  $R^{12}(R^{13})_a$ ; wherein:

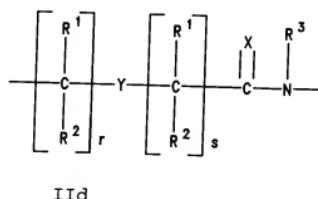
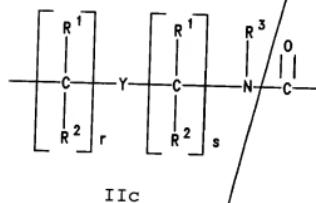
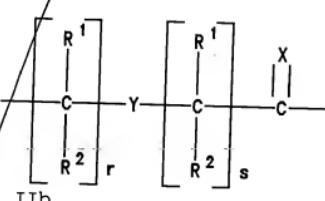
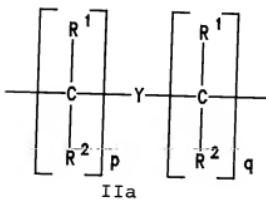
10        $R^{12}$  is hydrogen, hydroxy, ( $C_1-C_4$ ) alkanoyl, a naturally occurring nucleobase, a non-naturally occurring nucleobase, an aromatic moiety, a DNA intercalator, a nucleobase-binding group, a heterocyclic moiety, a reporter ligand, or a conjugate and at least one of  $R^{12}$  is a naturally occurring nucleobase, a non-naturally occurring nucleobase, a DNA intercalator, or a nucleobase-binding group;

15        $R^{13}$  is a conjugate; and  
a is 0 or 1.

E

A and B are selected such that:

- (a) A is a group of formula (IIa), (IIb) or (IIc) and B is N or  $R^3N^+$ ; or
- (b) A is a group of formula (IId) and B is CH;



5 where:

X is O, S, Se,  $NR^3$ ,  $CH_2$  or  $C(CH_3)_2$ ;

Y is a single bond, O, S or  $NR^4$ ;

p and q independently are zero or an integer from 1 to 5, the sum p+q being not more than 10;

10 r and s independently are zero or an integer from 1 to 5, the sum r+s being not more than 10;

$R^1$  and  $R^2$  independently are hydrogen,  $(C_1-C_4)$  alkyl,

hydroxy-substituted ( $C_1-C_4$ )alkyl, alkoxy-substituted ( $C_1-C_4$ )alkyl, alkylthio-substituted ( $C_1-C_4$ )alkyl, hydroxy, alkoxy, alkylthio, amino, halogen or a conjugate;

10 C is  $(CR^6R^7)_y$ ;

D is  $(CR^6R^7)_z$ ; wherein:

R<sup>6</sup> and R<sup>7</sup> independently are hydrogen, a side chain of a naturally occurring alpha amino acid, ( $C_2-C_6$ ) alkyl, aryl, aralkyl, heteroaryl, hydroxy, ( $C_1-C_6$ ) alkoxy, ( $C_1-C_6$ ) alkylthio, a conjugate, NR<sup>3</sup>R<sup>4</sup> and SR<sup>5</sup> or R<sup>6</sup> and R<sup>7</sup> taken together complete an alicyclic or heterocyclic system;

15 R<sup>3</sup> and R<sup>4</sup> independently are hydrogen, a conjugate, ( $C_1-C_4$ )alkyl, hydroxy- or alkoxy- or alkylthio-substituted ( $C_1-C_4$ )alkyl, hydroxy, alkoxy, alkylthio or amino; and

R<sup>5</sup> is hydrogen, a conjugate, ( $C_1-C_6$ )alkyl, hydroxy-, alkoxy-, or alkylthio- substituted ( $C_1-C_6$ )alkyl; each of y and z is zero or an integer from 1 to 10, the sum y + z being greater than 2 but not more than 10;

20 E independently is COOH, CSOH, SOOH, SO<sub>2</sub>OH or an activated or protected derivative thereof;

F independently is NHR<sup>3</sup> or NPgR<sup>3</sup>, where Pg is an amino protecting group; and

25 at least one of R<sup>1</sup>, R<sup>2</sup>, R<sup>3</sup>, R<sup>4</sup>, R<sup>5</sup>, R<sup>6</sup>, R<sup>7</sup>, R<sup>12</sup>, and R<sup>13</sup> is a conjugate wherein said conjugate is a reporter enzyme, a reporter molecule, a steroid, a carbohydrate, a terpene, a peptide, a protein, an aromatic lipophilic molecule, a non aromatic lipophilic molecule, a phospholipid, an intercalator, a cell receptor binding molecule, a

30 crosslinking agent, a water soluble vitamin, a lipid soluble vitamin, an RNA cleaving complex, a metal chelator, a porphyrin an alkylator, or a polymeric compound selected from polymeric amines, polymeric glycols and polyethers; and wherein said conjugate optionally includes a linking moiety.

B F E 223  
22. A peptide nucleic acid conjugate of claim 21  
wherein said conjugate includes a linking moiety.

B F Sub 223  
23. A peptide nucleic acid conjugate of claim 21  
wherein R<sup>12</sup> is a conjugate.

B F Sub 223  
24. A peptide nucleic acid conjugate of claim 21  
wherein R<sup>13</sup> is a conjugate.

B 223  
25. A peptide nucleic acid conjugate of claim 21  
wherein at least one group R<sup>3</sup> is a conjugate.

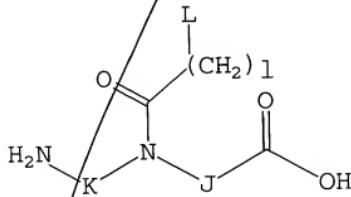
B 223  
26. A peptide nucleic acid conjugate of claim 21  
10 wherein at least one of said groups A or said groups B  
include a conjugate.

B 223  
27. A peptide nucleic acid conjugate of claim 21  
wherein at least one of group R<sup>1</sup> or group R<sup>2</sup> is a conjugate.

B 223  
28. A peptide nucleic acid conjugate of claim 21  
15 wherein at least one of R<sup>3</sup>, R<sup>4</sup>, R<sup>5</sup>, R<sup>6</sup>, and R<sup>7</sup> is a conjugate.

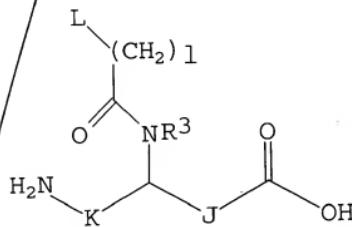
B 223  
29. A peptide nucleic acid conjugate of claim 21  
wherein at least one of said groups C or said groups D  
include a conjugate.

30. A peptide nucleic acid conjugate comprising a plurality of PNA monomers wherein at least one of said PNA monomers has the formula:

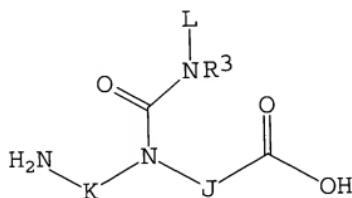


or formula:

5.



or formula:



225  
183

E wherein:

5 L is  $R^{12}(R^{13})_a$ ; wherein:

R<sup>12</sup> is hydrogen, hydroxy, ( $C_1-C_4$ ) alkanoyl, a naturally occurring nucleobase, a non-naturally occurring nucleobase, an aromatic moiety, a DNA intercalator, a nucleobase-binding group, a heterocyclic moiety, a reporter ligand, or a conjugate and at least one of R<sup>12</sup> is a naturally occurring nucleobase, a non-naturally occurring nucleobase, a DNA intercalator, or a nucleobase-binding group;

10 R<sup>13</sup> is a conjugate; and  
a is 0 or 1;

K is  $(CR^6R^7)_z$ ;

15 J is  $(CR^6R^7)_y$ ; wherein:

20 R<sup>6</sup> and R<sup>7</sup> are independently hydrogen, a side chain of a naturally occurring alpha amino acid, ( $C_2-C_6$ ) alkyl, aryl, aralkyl, heteroaryl, hydroxy, ( $C_1-C_6$ ) alkoxy, ( $C_1-C_6$ ) alkylthio, a conjugate, NR<sup>3</sup>R<sup>4</sup> and SR<sup>5</sup> or R<sup>6</sup> and R<sup>7</sup> taken together complete an alicyclic or heterocyclic system;

25 R<sup>3</sup> and R<sup>4</sup> independently are hydrogen, a conjugate, ( $C_1-C_4$ ) alkyl, hydroxy- or alkoxy- or alkylthio-substituted ( $C_1-C_4$ ) alkyl, hydroxy, alkoxy, alkylthio or amino;

30 R<sup>5</sup> is hydrogen, a conjugate, ( $C_1-C_6$ ) alkyl, hydroxy-, alkoxy-, or alkylthio- substituted ( $C_1-C_6$ ) alkyl; each of y and z is zero or an integer from 1 to 10, the sum y + z being greater than 2 but not more than 10;

35 1 is an integer from 1 to 5; and

at least one of R<sup>1</sup>, R<sup>2</sup>, R<sup>3</sup>, R<sup>4</sup>, R<sup>5</sup>, R<sup>6</sup>, R<sup>7</sup>, R<sup>12</sup>, and R<sup>13</sup> is a conjugate wherein said conjugate is a reporter enzyme, a reporter molecule, a steroid, a carbohydrate, a terpene, a peptide, a protein, an aromatic lipophilic molecule, a non aromatic lipophilic molecule, a phospholipid, an

5  
*fuller cont*  
intercalator, a cell receptor binding molecule, a crosslinking agent, a water soluble vitamin, a lipid soluble vitamin, an RNA cleaving complex, a metal chelator, a porphyrin an alkylator, or a polymeric compound selected from polymeric amines, polymeric glycols and polyethers, and wherein said conjugate optionally includes a linking moiety.

31. A peptide nucleic acid conjugate of claim 30 wherein said conjugate includes a linking moiety.

10 F 32. A peptide nucleic acid conjugate of claim 30 wherein ~~R<sup>12</sup> is~~ <sup>is 13</sup> a conjugate.

G 50 33. A peptide nucleic acid conjugate of claim 30 wherein R<sup>12</sup> is a conjugate.

15 34. A peptide nucleic acid conjugate of claim 30 wherein at least one of R<sup>3</sup>, R<sup>4</sup>, R<sup>5</sup>, R<sup>6</sup>, and R<sup>7</sup> is a conjugate.

35. A peptide nucleic acid conjugate of claim 30 wherein at least one of said group K or said group J includes a conjugate.

20 36. A peptide nucleic acid conjugate of claim 30 wherein said group R<sup>3</sup> is a conjugate.

Add B' > cold E 27

cold H 17 >  
Add J 13 >